ON TWO REMARKABLE SPOROCYSTS OCCURRING IN *MYTILUS LATUS*, ON THE COAST OF NEW ZEALAND.

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(Plates xix.-xx.)

The parasites which form the subject of the present communication were found by me last summer in specimens of a species of Mytilus (M. latus) living on the coast of New Zealand. I am greatly indebted to Dr. Chas. Chilton, Acting Professor of Biology at Canterbury College, Christchurch, for subsequently procuring and sending me a large consignment of live specimens of the mussel.

THE SPOROCYSTS OF AN ECHINOSTOMUM.

The Sporocysts, in which the Cercariæ are developed, were found in a fairly large proportion (about 10 per cent.) of the mussels examined, and always in large numbers. They infest the mantle-folds and the region immediately in front of the posterior adductor muscle, and are most abundant in the gonads and nephridia. In infected mussels this region of the body is coloured bright red owing to the presence of hundreds of the Sporocysts, which contain a conspicuous red substance to be referred to below.

From the fact that such large numbers of the Sporocysts occur together, it is to be inferred that they multiply actively in the tissues of the mussel. This multiplication of Sporocysts takes place not only by budding, or rather binary fission, but also, though comparatively rarely, by a process corresponding to that by which in many, if not most, Sporocysts Rediæ are developed. The Sporocysts (figs. 1 and 2) reach a maximum length of about 3 mm. They are able to creep about by alternate elongations and contractions. During these movements the shape necessarily varies greatly, but a "head," directed forwards, in locomotion, is always recognisable, being constricted off as a rounded knob. As Braun (3) points out, it is erroneous to refer to this as a sucker. The muscular layers in this position are not thickened or modified in any way. At its apex this is capable of being deeply involuted, the result being the development of a pit with a terminal opening. Since it is usually in this position that the fully developed Cercaria escapes, an actual perforation sometimes is found to occur, but this is of an entirely temporary character.

In many cases, applied to the outer surface of the Sporocyst were groups of amœboid cells emitting pseudopodia. Such groups of cells are stated by Biehringer to occur generally in the Sporocysts studied by him; the cells, which are blood-corpuscles of the host, eventually giving rise to a layer—"the paletot"— which completely invests the parasite. Such a continuous layer I have never found. The groups of cells were never found in entire preserved specimens or in sections, having always become detached, apparently, in the process of preparation.

The Sporocyst is enclosed in a non-cellular cuticular layer in which no trace of nuclei occurs. This layer, which is 0.003 mm. in thickness, exhibits a fine vertical striation, the significance of which will be referred to presently. Beneath it are the layers of muscle, which are extremely thin, an outer layer of circularly and an inner of longitudinally running fibres.

The outer layer (cuticle of most authors) is described as a thin structureless membrane. Biehringer (2), Heckert (*teste* Braun) and Loos (9) describe it as containing, in the young condition, sparsely distributed nuclei. In the Sporocyst from *Mytilus latus* nuclei were not found in the cuticle of any of the numerous specimens which were examined for them: it is possible, however, that the same may not hold good of those Sporocysts that are developed directly from the Miracidium embryo. In the Sporocyst of *Cercaria armata*, also, nuclei are absent in the cuticle according to Schwarze (12).

The germinal epithelium (fig. 8) lines the whole interior of the Sporocyst. In the greater part of its extent in a mature Sporocyst it is a thin layer of flattened cells, most of which are stellate, the processes of neighbouring cells anastomosing to form a network. At the anterior and posterior ends it is thickened, the thickening consisting of a layer of vertically elongated cells. At its outer end each cell of these thickened regions terminates in one or several very fine processes, which are clearly traceable through the layer of muscle, and are seen to be continuous with the vertical striations of the cuticle. At its opposite free end each cell projects into the cavity of the Sporocyst. In the living specimen globules were seen to be given off occasionally from the inner ends of these cells, becoming free in the interior of the cavity, and often becoming collected in masses. These globules were swallowed by the more mature Cercariæ, in the intestine of which considerable numbers of them were nearly always to be detected. It might be supposed that this phenomenon is not a natural one, but brought about by the pressure to which the living Sporocyst is naturally subjected in order that its structure may be examined. But such an explanation cannot be entertained in view of the results obtained from the examination of sections of specimens fixed by various reagents. In such sections the epithelium has the appearance represented in in fig. 8, the cells presenting a variety of stages in the process of formation of the globules; and many of the latter are to be found free in the cavity of the Sporocyst and in the intestine of the Cercariæ. Among the globules in the latter position entire nucleated cells are frequently to be observed.

It thus appears to be almost certain that the germinal epithelium has an important secondary function in providing nutriment for the Cercariæ with fully developed and functional digestive canal. To nourish the many growing embryos which it contains, the wall of the Sporocyst must absorb nutrient matter in large quantity from the tissues of its host. This is taken in, undoubtedly, by the cells of the germinal epithelium through the pores of the cuticle, and most of it must be given off in a state of solution into the fluid in which the developing embryos, in all but their earliest stages, float freely. The special part played by the nutriment provided in the form of globules is that it supplies a store of sustenance which the Cercaria takes with it when it leaves the Sporocyst and sets out in search of a second host.

I can find no mention in the literature accessible to me of this giving off of globules by the cells of the germinal epithelium. That the case is not an altogether isolated one, however, is shewn by the fact that Reuss in a recently published preliminary notice (11) mentions the presence in the Sporocysts of *Distomum duplicatum* of many "fat drops." It is at least possible that the phenomenon is a general one, and that it is only owing to the colourless character of the globules in most instances that it has hitherto escaped notice. On the other hand, it has to be borne in mind that in many Cercariæ the digestive canal does not become sufficiently well developed to be functional until after the larva has escaped from the Redia or Sporocyst. In such cases the formation of the globules, except merely in order to be subsequently dissolved in the nutrient fluid of the cyst, would evidently be superfluous.

The occurrence of a pigment in the germinal epithelium appears to be exceptional. I have met with only three recorded cases of such a thing—by Wagener (14) in the Sporocyst of *Cercaria cystophora*, by Pagenstecher (10) in the case of the Sporocyst of *Cercaria cotylocerca*, and by Heckert (4) in that of *Leucochloridium paradoxum*.

The embryos are developed from a mass of specialised cells of the germinal epithelium—the ovary (figs. 5-7). This is constant in position, at the posterior end, and there is never more than one. It projects as a free process into the cavity of the Sporocyst. At its base every gradation is to be observed between the ordinary cells of the posterior thickening of the germinal epithelium and the cells destined to give rise to embryos. Towards its free extremity the ova undergo segmentation which results in

the formation of a spherical body containing a number of nuclei with cell-outlines ill defined, though sometimes recognisable. These spherical embryos are still enclosed within a delicate membrane which invests the whole ovary. As they reach a size of 0.025 mm. or thereabouts, they become free, and eventually develop into the mature Cercaria condition while floating in the fluid in the interior of the Sporocyst.

In Amphistomum subclavatum according to Loos (9), in Leucochloridium paradoxum according to Heckert (4), and in Distomum duplicatum according to Reuss (11), the formation of the ova originally takes place at any point in the wall of the Sporocyst a definite ovary only becoming established at a later stage. Whether this may hold good in the case of the Sporocyst from Mytilus latus with regard to the primary Sporocyst developed from the Miracidium there are no data from which to decide. I have found no Sporocysts, however small, among the hundreds examined in which the formation of ova was not localised in a single ovary; but I may not have seen any except those formed secondarily by fission or endogenous formation, or at all events may not have happened to meet with any early primary Sporocysts.

The cells of the germinal epithelium give rise, however, not only to embryo Cercariæ, but sometimes also, though very rarely so far as my specimens are concerned, to a new generation of Sporocysts. These become set free in the interior of the parent Sporocyst. From their germinal epithelium, before they escape to the exterior, embryos of Cercariæ may already have become formed. The young Sporocysts (fig. 3) on escaping readily multiply by binary fission (fig. 4): a constriction appears dividing the Sporocyst into two equal or slightly unequal parts, and by the deepening of the constriction the two parts become eventually completely severed.

In young Sporocysts the germinal epithelium is of uniforn[†] thickness throughout; but at a very early stage an anterior extremity capable of involution becomes differentiated, after which, the cyst increasing in length, the germinal epithelium soon comes to be attenuated except at the anterior and posterior extremities. In young Sporocysts the internal cavity, owing to the small size of the cyst and the thickness of the germinal epithelium, is extremely small. Occasionally in sections of later as well as earlier stages a delicate reticulated substance appears to fill this cavity wholly or partially: but there can be no doubt that, at least so far as the later stages are concerned, this is an entirely artificial product formed as a result of the coagulation of the fluid contained in the cyst.

THE MATURE CERCARIÆ.

The fully developed Cercaria (fig. 9) reaches a total length of about 4 mm., the tail being included. The head or part of the body bearing the anterior sucker is quite distinctly marked off from the rest, being much narrower. On its ventral aspect is the large cup-shaped anterior sucker, which is capable of being completely everted. Around the anterior margin of the head on the dorsal side, and usually inclined outwards and backwards, are a series of flattened tooth-like spines which are capable of being turned forwards. Eyes are not present. At the sides of the head region are a series of six nearly transverse ridges which do not extend on to the dorsal or ventral surfaces. The remainder of the body is of oval, dorso-ventrally compressed, form; in front of the middle is the posterior sucker.

The tail consists of a main trunk and two branches. When extended to its utmost the former is over three times the length of the body; the branches are nearly as long as the trunk. The latter expands proximally where it joins the body into a disc, the margin of which is divided by notches and fissures into a series of branched lobes. I have not seen this peculiar structure performing any function; but it can hardly be anything else than an organ of adhesion. It contracts and expands slowly at irregular intervals. The main axis of the tail, when at rest, is not in line with the body, but bent towards the ventral side. It performs extremely rapid vibrating movements, propelling the larva swiftly through the water. The tail has a remarkable segmented appearance, due to the presence of the regularly arranged transverse bundles of muscular fibres referred to below.

The body is enclosed in a cuticle 0.003 mm. in thickness. Beneath this is a muscular layer consisting of external, longitudinal, and internal circular fibres. The space between the body-wall and the enteric canal is filled with parenchyma, in which are numerous large cells about 0.04 mm. in diameter — the cystogenic cells. In young specimens these contain a coarse reticulum. In mature specimens they appear more homogeneous. Their ducts (processes of the cells) branch, and the branch ducts perforate the cuticle to open over the entire surface of the body. There are no cystogenic cells in the tail. No rhabdites were observed in any part.

The pharynx (fig. 10) is a relatively long cylinder. The region of the digestive canal following upon this behind, commonly known as the œsophagus, is very short. There is nothing special to be noted with regard to the structure of the various parts of the digestive system. The intestine is fully developed long before the Cercaria becomes mature.

The tail (figs. 11 and 12) is a hollow cylinder with a thin wall, tensely filled with a watery fluid. It has a thin cuticle, beneath which is a single layer of circular muscular fibres of extreme Internal to this are four bundles of longitudinal fineness. muscular fibres, two dorsal and two ventral. On each side is a fairly wide interval-the *lateral line* as it may conveniently be termed—between the dorsal and the ventral bundle. Between the dorsal and ventral bundles on each side run a number of vertical (dorso-ventral) fibres for the most part in narrow strands occasionally with nuclei. These are arranged at intervals along the tail in such a way as to produce in some cases an appearance of segmentation. Along each lateral line, immediately below the cuticle and circular muscle, runs a delicate filament. This gives off branches which run both dorsally and ventrally, frequently dividing into finer threads. At several points in its course the main filament presents a thickening. Accompanying this is a very regular row of small, usually rounded, cells of a peculiarly

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clear and bright appearance in the living condition, so that they are very conspicuous even under a low power. These, which are separated from one another by very regular intervals, are all connected with the lateral filament, usually by means of sidebranches, sometimes (rarely) directly with the main filament itself, which then seems to pass through them. There is no direct evidence of the nature of these filaments of the lateral line; but from the way in which they branch I have very little doubt that they are nerves. If this should prove to be correct, the regularlyarranged cells connected with them, though they have no processes projecting on the exterior, are probably sense-cells of some sort.

Applied to the inner surfaces of the bundles of longitudinal muscular fibres are a number of very large granular cells about 0.04 mm. in diameter, with vesicular nuclei 0.01 mm. in diameter. As shown by their relations and their developmental history these are the myoblasts of the longitudinal muscular fibres.

The branches of the tail resemble the tail proper in structure, except that the transverse fibres are more irregularly arranged and closer together. In the lobed disc at the proximal end of the trunk the bundles of longitudinal fibres become spread out so as to be inserted into the lobes. The dorso-ventral bundles are here represented by thick columnar cells.

Not much attention appears to have been paid to the structure of the tail in Cercaria; but, so far as I am able to judge, the form at present under consideration would appear to be exceptional in this respect, especially with regard to the entire absence of any axial strand. Thus Leuckart states (6, p. 512): "Unterhalb der Cuticula erkennt man deutliche Rings- und Längsmuskeln und das nicht blos am Rumpfe, sondern auch am Schwanze, nur dass sich letztere hier gewöhnlich rechts und links zu breiten Bändern gruppiren, und somit eine Anordnung zeigen die man bei aufmerksamer Betrachtung schon aus der entschieden seitlichen Bewegungen des Schwanzes erschliessen könnte. Die Zellen des Achsenstranges sind in eine glashelle Bindesubstanz verwandelt die nur noch einzelne spindelförmige Körper erkennen lässt und nach Art eines (unvollständig erhärteten) Skelettes dazu dienen

möchte, den Schwanz elastisch zu machen und ihm einen gewissen Grad von Rigidität zu verleihen. Zwischen Muskeln und Achsenstrang liegt eine Schicht von grossen platten Zellen, die bei Cercaria macrocerca bei welcher der Achsenstrang zu eines mächtigen Entwickelung heranwächst, sehr sonderbar verästelte Formen haben."

Here it will be observed the axial strand is described as being composed of cells, which have become converted into a hyaline connective tissue acting as the main support of the organ.

The structure of the tail in *Bucephalus* is described by Ziegler (18), but as this is by no means a typical Cercaria the statements which he makes probably do not hold good generally; they are certainly entirely inapplicable to the form which I am now describing.

Thiry (13) states that in *Cercaria macrocerca* the tail has an external circular layer of muscle. The longitudinal fibres run in two broad bands at the sides. Beneath this is a mass which anteriorly consists of large cells, posteriorly appears homogeneous.

Schwarze's account (12) of the structure of the tail in *Cercaria* armata is also not in any way in agreement with what is to be observed in the Cercaria from *Mytilus*. He describes the contractile substance of the tail as forming an axial strand surrounded by a layer of cystogenic cells, and these again by a further layer of contractile substance, the outer layer being connected with the axial strand by numerous radiating bundles.

Reuss (11, p. 378) states that in the Sporocyst of *Distomum* duplicatum the tail has a layer of cells underlying the cuticle, and the interior is traversed by transverse muscular fibres.

It does not seem to be possible to reconcile these various statements, and, if they are correct, there must be considerable diversity in the structure of the tail in different Cercariæ.

In the posterior part of the body opening near the root of the tail is the excretory bladder. This is broad behind, narrower in front, where it becomes forked. Each of the branches terminates anteriorly in a slight enlargement into which opens the corresponding main excretory vessel, right or left as the case may be. Each of these runs outwards and forwards to a point near the margin of the body, where it divides into an anterior and a posterior longitudinal trunk. The ciliated funnels are 0.0075 mm. in diameter. There are always twelve of them in mature Cercariæ, and their arrangement is quite constant. One funnel terminates each of the anterior and posterior longitudinal vessels; the remainder are situated at the ends of longer or shorter sidebranches in the way shown in fig. 10.

No funnels and no vessels were observed in the tail. In this respect this Cercaria differs apparently from other described forms. Wagener (17) states that in the Cercariæ with forked tail the excretory bladder extends into the tail and becomes bifurcate, each branch opening at the end of the corresponding branch of the tail; and Loos (7) describes and figures the extension into the tail of the main excretory vessel in the Cercariæ of Amphistomum subclavatum.

The reproductive system is fairly well advanced in the mature Cercaria. The ovary is a rounded mass of cells situated a little behind the point of bifurcation of the intestine. From it the oviduct proceeds as a narrow tube which runs forwards and towards the left passing on the ventral side of the left ramus of the intestine, and bending in again towards the middle line to open at the genital aperture immediately in front of the ventral sucker. A rounded mass surrounding the oviduct shortly after it leaves the ovary is evidently the shell-gland. The rudiments of the intestine some distance behind the ovary. The vitelline glands are not yet distinguishable. A rounded body dorsal to the anterior part of the ventral sucker is probably the vesicula seminalis.

In the form and arrangement of the spines around the anterior end, the series of lateral ridges in the region immediately following, and the form of the pharynx, this Cercaria resembles the adult *Distomum fasciatum*, Rud., and probably is the larva of a form nearly related to that species.* In the first point it also resembles *D. acanthocephalum* and other members of Rudolphi's sub-genus *Echinostomum*.

DEVELOPMENT OF THE CERCARIÆ.

For what we know of the development of the Cercaria within the Sporocyst or Redia we are mainly indebted to Leuckart (17), Thomas (14), Heckert, Schwarze (12), and Loos (9). In spite of their labours, however, it must be said that a good many points remain obscure. This holds good particularly with regard to the precise mode of origin of certain of the organs; and it is due mainly to the almost complete absence of differentiation among the cells (meristem cells of Schwarze and others) of the early stages, the rudiments of various parts having the appearance of becoming quite suddenly crystallised out from a previously homogeneous mass.

As already stated, the embryos of the Cercaria from Mytiluslatus are set free from the ovary when they have attained a diameter of about 0.025 mm.* At about this stage (figs. 15 and 16) the embryonic investment is formed. First a single cell on the surface becomes flattened out, then others become similarly modified until the spherical mass of cells becomes enclosed in a

* Reuss (11) met frequently with a stage in which one large cell (ovum) was accompanied by three considerably smaller cells, with nuclei 0.0025 in diameter with uniformly distributed, coarsely granular chromatin. Since the large cell next divides into quite equal and similar cells he takes these previously formed three smaller cells to be of the nature of polar bodies, and their formation to be a maturation process. This stage I have not observed. But in a large proportion of specimens there occur lying loose in the Sporocyst in the immediate neighbourhood of the ovary a varying number of cells (fig. 14) which have homogeneous deeply staining nuclei 0.002 mm. in diameter. If these are not of the nature of polar bodies it seems difficult to account for them.

^{*} I infer this from Braun's reproduction (3, taf. xxi., fig. 8) of a figure by Stossich, the original paper by that author (in the Bolletino della Soc. adriat. scienze natur. Trieste, vol ix., 1885) not being accessible to me.

thin layer composed of a small number of flattened cells which become completely united at their edges. This investment persists to a comparatively late stage in the development of the Cercaria, only disappearing as the cuticle becomes formed.

Schwarze's account (12) of the development of the investing layer in *Cercaria armata* agrees closely with what I have observed. I cannot, however, follow him in his opinion that the embryonic investment gives rise to the cuticle of the Cercaria. The investment in question is an excessively thin membrane which at no time contains more than about half-a-dozen nuclei. As the embryo grows, this simply becomes stretched, and in early tailed stages appears quite loosely connected with the underlying layers. I have very little doubt that it is the layer of cells which, at the period when the tail is being formed, becomes arranged in a regular manner beneath the original investing layer, that secretes the cuticle.

Heckert^{*} states that in embryo Cercariæ of a diameter of 0.075 mm. a second investment similar to the first appears beneath it; and Loos (9) observed a similar phenomenon in the development of Rediæ (letter to Braun quoted in foot-note, 3, p. 818). There is no appearance of this second investing layer in the case of the Cercaria from *Mytilus latus*.

Small nuclei with deeply-staining, apparently homogeneous, plasma, similar to those described by Schwarze (10) in *Cercaria armata* as giving rise to the rudiment of the reproductive system, appear before the embryo leaves the ovary. But these have no definite arrangement, occurring, frequently two together, here and there, both near the surface and more deeply, instead of forming a central mass as stated by Schwarze. They are most probably merely phases in the history of the ordinary meristem nuclei, and have no special histogenetic significance.

^{*} I only know this paper ('Untersuchungen über die Entwickelungs- und Lebensgeschichte des Distomum macrostomum.' 'Bibliotheca Zoologica,' Leuckart u. Chun, Heft 4), as quoted by Braun (3).

As the embryo increases in size, it becomes somewhat oval, and soon, when the long diameter is about 0.08 mm., the future posterior end becomes marked by a slight depression, the anterior end remaining rounded (fig. 17). In this position a narrow cleft appears running forwards towards the centre of the embryo. This cleft does not open on the exterior, but is covered over by the thin investing layer of cells. On either side of it is a mass of cells from which the tail buds out; in front it terminates in a slight transverse enlargement which partly separates off a dense central mass of cells from two lateral masses. The enlargement subsequently becomes the bladder of the excretory system. In the lateral masses the rami of the intestine are developed at a later stage.

The central mass of cells is distinguishable (figs. 18 and 19) into an anterior and a posterior portion which are not sharply marked off from one another. The posterior portion is the rudiment of the reproductive system. Of the anterior portion the more ventrally placed cells form the rudiment of the pharynx. the more dorsally placed that of the brain. Of these two organs the pharynx appears first as a cylindrical cord of cells soon distinguishable (fig. 20) into an investing layer and a central core. The cells of the investing layer are the myoblasts of the wall of the organ. They become enlarged, and increase in number, though never becoming very numerous. In their plasma are developed the radial muscular fibres of the pharynx. The enclosed row of cells do not persist. They at first fill the entire lumen, but spaces appear, and the cells gradually become absorbed and eventually entirely disappear (fig. 22). Whether they give rise to the thin cuticle lining the interior of the pharynx is uncertain. It is difficult to understand by what other agency that membrane could be formed.

At the anterior end, towards the ventral surface, when the rudiment of the pharynx first becomes formed, a rounded groupof cells in all respects similar to the rest becomes enclosed by a thin membrane formed of flattened cells. This is the rudiment of the anterior sucker. A continuation backwards of the cord of cells which forms the rudiment of the pharynx constitutes the rudiment of the short œsophagus. This soon bifurcates posteriorly to form the biramous intestine. In the latter the central cells of the cord become enclosed, as in the case of the pharynx, in a cylinder formed of the peripheral cells. But the latter layer remains very thin, developing into the thin layer of muscle of the wall of the intestine, while the enclosed cells, few and irregularly disposed at first (fig. 23), multiply rapidly, at the same time becoming disposed as a regular epithelial layer (figs. 24-26) bounding the lumen. In the œsophagus the muscular investing layer also remains thin, but the enclosed cells give rise to a layer devoid of nuclei, but divided into numerous minute bodies somewhat smaller than the cells of the epithelium of the intestine.

In *Cercaria armata*, Schwarze does not refer to the formation of a cleft at the anterior end preceding the development of the unpaired part of the enteric cavity; but describes the latter as appearing in the form of a solid process which only develops a lumen at a later stage by the absorption of the axial cells. In that Cercaria the full development of the forked intestine only takes place after the Cercaria has become encysted, and the entire enteric canal can be functional in the Cercaria stage only very imperfectly, if at all.

The rudiment of the posterior sucker is distinguishable as a rounded projection on the veutral surface, shortly after the tail begins to be developed; this consists, like the anterior sucker, of a rounded mass of undifferentiated cells separated internally from the rest of the body cells by a thin membrane.

When the rami of the intestine first make their appearance, a space—the origin of which has been already referred to—is developed between them. This is the median bladder of the excretory system. At first it has no definite wall, but, later, cells become arranged to form a thin epithelium as in the adult.

When the cord of cells destined to give rise to the pharynx first becomes differentiated, a mass of cells, as already stated, lies on the dorsal side of it. From the central part of this the first rudiment of the nervous system becomes formed. This appears as a bridge of finely fibrillated material (fig. 27) running transversely over the pharynx and surrounded by numerous cells—the rudiment of the brain. This is well established when the pharynx is still a narrow tube with a thin wall enclosing a core of cells. From the brain two narrower strands of similar material run backwards at the sides, each surrounded by groups of cells; and are traceable for some distance through the body, though not extending at first very far back. Before long each longitudinal cord bifurcates to form a dorsal and a ventral branch. About the same time a nerve is formed running forwards from the brain towards the anterior extremity.

The tail (figs. 29-31) appears first, when the embryo is about 0.09 mm. in length, as a rounded process consisting of an undifferentiated mass of cells covered by a continuation of the thin investing layer. While still very short it becomes bilobed at the extremity. Before this takes place internal differentiation has begun. A very regularly arranged line of small nuclei becomes ranged along each side below the investing layer; and the rest of the tissue forms a central core. As the two branches grow out at the bilobed extremity, their constituent cells take on a similar arrangement. The central core becomes divided into four longitudinal rows of cells-two dorsal and two ventral; these are the myoblasts. They become enlarged, and their nuclei become vesicular. In the substance of their protoplasm external to the nuclei appear bright-looking longitudinal bands which stain deeply with eosin; these are the first-formed muscular fibres. They soon become more numerous, and as the tail becomes fully developed, take the form of the four longitudinal bands. The small cells of the lateral rows, at first placed close together, become separated from one another by distinct intervals as the tail elongates, and become the lateral series of small cells referred to above in the account of the tail of the adult.

THE SPOROCYSTS AND CERCARLÆ OF A SPECIES OF GASTEROSTOMUM.

Only one specimen of the mussel was observed to be infested with this parasite. It presented a very remarkable appearance. The mantle-folds and the visceral mass in the region between the adductor muscles had the appearance of being provided with a ramifying system of vessels containing a blood-red fluid. These apparent vessels, when examined under the microscope, were found to be narrow ramifying Sporocysts, the germinal epithelium of which contained a red colouring matter apparently identical with that occurring in the Sporocyst described in the first part of this paper.* In these Sporocysts were mature Cercariae and all stages in their development. The Cercariae proved to belong to the remarkable form known as *Bucephalus*, v. Baer, the larva of *Gasterostomum*. All of these are developed in narrow, tubular, usually branching Sporocysts; but in no case hitherto recorded, so far as I am aware, is a red colouring matter present.

The Cercariæ (fig. 32) are relatively small, being only 0.25 mm. in length when fully matured. They are sluggish in their movements, and the tail was never observed to be used as a swimming appendage, its only movements being slow waving ones, or such as resulted in each of the branches being coiled into a spiral and uncoiled again. Such spiral movements were noticed by v. Baer in *Bucephalus polymorphus*. It seems likely that the tail is used more for attachment than as an organ of active locomotion.

The body is long and narrow. The anterior portion, lodging the anterior sucker or proboscis, is separated off from the rest by a slight constriction. Its anterior end (figs. 33-36) varies in appearance according to its condition. When somewhat contracted it usually appears trilobed; when it is more extended the rounded aperture of the sucker appears at its extremity. The anterior extremity is beset with excessively minute cuticular spinules which gradually decrease in size as they pass backwards, and are no longer distinguishable when the middle of the body is reached. A pair of relatively long and slender cilia, presumably sensory, extend forwards from the extreme anterior end.

^{*} The nature of this colouring matter has not yet been investigated. Its spectum does not shew the absorption bands characteristic of that of hæmoglobin.

The mouth is situated on the ventral surface considerably behind the middle. It leads into a rounded pharynx, from which the simple intestine leads forwards as far as, or further than, the middle of the length of the body. No contents of any kind were ever observed. Behind the mouth and pharynx are two irregular masses of cells, forming a slightly unsymmetrical pair; these are apparently rudiments of the gonads. Close to them in the middle is the excretory sac.

The tail consists of a basal part and two long branches or appendages. The former is somewhat elliptical, with the long axis transverse, the anterior border nearly straight, or at least less convex than the posterior. The branches are attached to the basal part antero-laterally; they are slender, nearly twice the length of the body, and taper slightly at the extremity.

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EXPLANATION OF PLATES.

Reference Letters.

br., brain. c., cuticle. cd., disc at base of tail. d.v.m., dorso-ventral muscles of tail. ex., principal excretory vessel. ex.b., excretory bladder. i., intestine. l.c., lateral cells of tail. l.m., longitudinal muscles of tail. l.n., lateral nerve of tail. m., mouth. m.f., muscular fibres developing in myoblasts. my., myoblasts. o.d., oviduct. œ., œsophagus. ov., ovary. ph., pharynx. r., rudiment of gonads. s'., anterior sucker. s"., posterior sucker. sp., spines. te., testes.

Plate xix.

- Fig. 1.—Sporocyst from Mytilus latus, magnified.
- Fig. 2.—Sporocyst, entire preserved and stained specimen, somewhat compressed (\times 75).
- Fig. 3.—Young stage of Sporocyst, compressed (\times 75).
- Fig. 4.—Young Sporocyst in the act of undergoing fission ($\times 75$).
- Fig. 5.—Ovary : appearance in entire specimen (\times 1000).
- Fig. 6.—Ovary: section, showing ova and early stages ($\times 1000$).
- Fig. 7.—Ovary: section next to that represented in fig. 6.
- Fig. 8.—Wall of Sporocyst: vertical section of one of the thickened regions of the germinal epithelium (×1000).
- Fig. 9.--Cercaria, magnified. The tail is here not so long in proportion as it is destined to become.
- Fig. 10.—Body of Cercaria (with base of tail), showing excretory and digestive systems. The posterior sucker is not shown.
- Fig. 11.—Transverse section of tail ($\times 750$).
- Fig. 12.—Lateral view of a portion of the tail, showing the lateral line and its nerve $(\times 340)$.

Fig. 13.—Two cystogenic cells (×950).

Fig. 14.—Cells supposed to be possibly of the nature of polar bodies (\times 1500)• Fig. 15.—Early free stage (\times 1000).

Plate xx.

Fig. 16.—Somewhat later stage than that represented in fig. 15 (\times 950).

Fig. 17.—Stage with flattened posterior end (\times 1000).

- Fig. 18.—Stage with clefts and rudiments of pharynx and reproductive system : entire specimen in optical section.
- Fig. 19.—Longitudinal section of similar stage: combined from two adjoining sections (×750).
- Fig. 20.—Transverse section of pharynx at the stage in which its wall has become well established but the musculature has not become formed (\times 700).

Fig. 21.—Transverse section of pharynx at a somewhat later stage : the cells still intact in the interior.

Fig. 22.—Somewhat later stage in which the cells in the lumen have disappeared and the cuticle has become developed (×700).

Fig. 23.—Transverse section of early Cercaria through the posterior region, showing the intestinal epithelium in the condition of irregularly distributed cells enclosed within the developing muscular layer (×700).

Fig. 24. — Transverse section of one of the rami of the intestine at the stage in which the internal cells, though still few in number, have become arranged in a definite layer (×1500).

- Fig. 25.—Section similar to that represented in fig. 24, but of a somewhat later stage, in which the epithelium has become more fully developed ($\times 1500$).
- Fig. 26.—Section of a small part of the epithelium of the intestine in the mature Cercaria, to show the form and arrangement of the cells $(\times 1500)$.
- Fig. 27. —Section through Cercaria in the region of the brain at a stage when muscular tissue is only beginning to be formed (\times 750).
- Fig. 28.—Tail at an early stage in its development: viewed as a transparent object (×750).

Fig. 29.-Similar view of later stage.

- Fig. 30.—Transverse section of tail at a stage corresponding to that represented in fig. 23 (\times 700).
- Fig. 31.—Transverse section of later stage in which the cuticle has become formed and the formation of the muscular fibres has begun.

Fig. 32.—Cercaria stage (*Bucephalus*) of a species of Echinostomum (×240). Figs. 33-36.—Outline of head end of living Bucephalus in different conditions.